REMARKS/ARGUMENTS

Status of the Application

In the Non-Final Office Action mailed June 30, 2006, claims 1-10, 14, and 16 were rejected. In the present response, claims 1-3, 7-10, 14, and 16 were amended to limit the carotenoid overproducing microorganism to *E. coli* (see canceled claim 6 for support), and claims 4-6 were canceled. Claim 3 was further amended to correct a grammatical error, and claim 16 was further amended to remove redundancies in the carotenoid list. Thus, claims 1-3, 7-10, 14, and 16 are pending. No new matter was added.

Rejections Under 35 U.S.C. § 112, 1st Paragraph

Claims 1-10, 14, and 16 were rejected under 35 U.S.C. § 112, 1st Paragraph, because the specification, while being enabling for an isolated β-carotene overproducing *Escherichia coli* host cell comprising the plasmid pPCB15 (cam^{R}) (SEQ ID NO:43) encoding the carotenoid biosynthesis gene cluster (*crtEXYIB*) from *Pantoea stewartii*, wherein *deaD* gene is disrupted, does not reasonably provide enablement for any carotenoid overproducing microorganism comprising genes encoding a functional isoprenoid enzymatic biosynthetic pathway comprising a disrupted *deaD* gene. Applicants respectfully traverse these rejections.

The touchstone of the enablement requirement is whether the skilled person can make and use the invention without undue experimentation. *In re Wands*, 858 F.2d 731, 737 (Fed. Cir. 1988). Applicants submit that the skilled person, in possession of the present application describing carotenoid overproducing *E. coli* having a functional isoprenoid enzymatic biosynthetic pathway and a disrupted *deaD* gene, in conjunction with well-known protocols of molecular biology (*see, e.g.*, page 29, lines 13-20, of the Specification), would have no difficulty in practicing the invention without undue experimentation.

As noted by the Examiner, the Wands factors are: (a) the quantity of experimentation necessary, (b) the amount of direction or guidance presented, (c) the presence or absence of a working example, (d) the nature of invention, (e) the state of prior art, (f) the relative skill of those in the art, (g) the predictability or unpredictability of the art, and (h) the breath of the claim. In relation thereto,

Applicants respectfully assert the following: Factor (a), the amount of experimentation needed to practice the invention is reasonable and commensurate with the art. Factor (b), the Specification provides description of a significant number of exemplary upper isoprenoid pathway genes, including endogenous *E. coli* upper isoprenoid pathway genes (*see* page 13, line 31 – page 18, line 2 of the Specification; especially Table 2), and a significant number of exemplary lower isoprenoid pathway genes (*see* page 18, line 4 – page 24, line 6; especially Table 3) available for construction of the isoprenoid pathway in *E. coli*. The specification also provides description of the disruption of the *E. coli deaD* gene (*see* page 24, line 7 – page 27, line 7 of the Specification). One skilled in the art should be familiar with the methods for *E. coli* gene transformation and disruption described in the Specification. Factor (c), Applicants have provided working examples demonstrating the production of an *E. coli* carotenoid overproducing microorganism, which has a functional isoprenoid pathway and a disrupted *deaD* gene. Factor (d), the invention is one of gene mutations in *E. coli*. Such art requires some experimentation for even routine techniques. Therefore, one skilled in the art would expect some experimentation, screening, and trial and error to implement the present invention outside the working examples. However, the information presented in the instant application is sufficient to enable one skilled in the art to implement the gene transformations and disruptions to practice the invention. Factor (e), as stated in the background of the invention (*see* page 1, line 35 – page 2, line 7 of the Specification), the carotenoid pigment biosynthesis pathway is extremely well-known. Previous attempts to increase carotenoid production in *E. coli* through genetic manipulation have focused on overexpression of isoprenoid pathway genes (*see* page 3, lines 1-13, of the Specification), which can lead to lethal accumulation of isoprenoid precursors (*see, e.g.*, Sandmann G, *Trends Plant Sci.* 6:14-17 (2001), cited at page 3, lines 25-26, of the Specification). Applicants have successfully developed carotenoid overproducing *E. coli* without direct manipulation of the isoprenoid pathway genes. Factor (f), as stated above, this invention is related to the biotechnical arts in an extremely well-known pathway, and the skill level of the artisan is very high. The skilled artisan is therefore very familiar with the pathway and well versed in many methods and techniques of gene manipulation. Factor (g), the biotechnical art is an

unpredictable art; it is not reasonable for an applicant to provide a cookbook recipe of how to practice the invention. Rather, Applicants have depended on the skill and experience of the artisan to implement the invention using the isoprenoid pathway genes of their choosing. It is expected that the artisan would be aware of successful methods of *E. coli* gene transformation and mutation and therefore be capable of implementing the described genetic manipulations in *E. coli*. Factor (h), the breath of the claim is reasonable given the vast improvement and the ability of skilled artisans to implement the invention into *E. coli*. It would be unfair to the Applicants to limit their invention to the working examples as the Specification has provided enough description to allow others in the art to use the present invention with *E. coli* having a functional isoprenoid enzymatic biosynthetic pathway and a disrupted *deaD* gene.

Claims 1-10, 14, and 16 were rejected under 35 U.S.C. § 112, 1st Paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventors, at the time the application was filed, had possession of the claimed invention.

The claims have been amended to recite the limitation that *E. coli* is the carotenoid overproducing microorganism. A carotenoid overproducing *E. coli* is well-described in the working examples and throughout the specification. Additionally, the source of the upper and lower isoprenoid pathway genes are well-described in the Specification (*see* page 13, line 31 – page 24, line 6; especially Tables 2 and 3), and disruption of the *E. coli deaD* gene is similarly well-described (*see* page 24, line 7 – page 27, line 7 of the Specification). The function of each gene is described in detail in the discussion entitled "Genes Involved in Carotenoid Production", beginning at page 13, line 14. Thus, the Specification provides a selection of species for each genus of isoprenoid pathway genes in the pathway, has provided a functional description of each gene, has pointed the skilled person to a number of specific sequences with structural features for each gene, and has fully described disruption of the *deaD* gene. Applicants thus respectfully submit that their Specification has put the skilled person on notice that Applicants were in possession of the invention at the time of filing.

SUMMARY

In view of the foregoing remarks, Applicants submit that this application is in condition for allowance. In order to expedite disposition of this case, the Examiner is invited to contact Applicants' representative at the telephone number listed below to resolve any remaining issues. Should there be a fee due which is not accounted for, please charge such fee to Deposit Account No. 04-1928 (E.I. du Pont de Nemours and Company).

Respectfully submitted,

By: /S. NEIL FELTHAM/
S. Neil Feltham
Attorney for Applicants
Reg. No.: 36,506
Telephone: (302) 992-6460
Facsimile: (302) 992-5374

Dated: 12/21/2006